

AMENDMENTS TO THE CLAIMS:

This listing of claims will replace all prior versions, and listings, of claims in the application:

Listing of Claims:

1-61. (Canceled)

62. (Previously presented) A composition comprising: an isolated Tat protein, fragment or mutant in combination with a pharmaceutically acceptable carrier or excipient, wherein said isolated Tat protein, fragment or mutant is biologically active, as shown by the ability of said isolated Tat protein, fragment or mutant to

- (i) become internalized by activated endothelial cells or dendritic cells, which internalization is determined by (a) incubating activated endothelial cells or dendritic cells with up to 1 $\mu\text{g/ml}$ of said isolated Tat protein, fragment or mutant which is labeled with rhodamine, and (b) detecting the presence or absence of rhodamine in the activated endothelial cells or dendritic cells by fluorescence microscopy; or
- (ii) activate the proliferation, migration, and invasion of Kaposi's sarcoma (KS) cells or cytokine-activated endothelial cells in culture when said Tat protein, fragment or mutant is present at a concentration of up to 1 $\mu\text{g/ml}$; or
- (iii) activate virus replication when said isolated Tat protein, fragment or mutant is added to HIV-1 infected cells at a concentration of up to 1 $\mu\text{g/ml}$, which activation is determined by (a) the rescue of Tat-defective proviruses in HLM-1 cells after the addition of said isolated Tat protein, fragment or mutant, or (b) the transactivation of HIV-1 gene expression in cells transfected with HIV-1 promoter-reporter plasmid,

wherein said composition is pharmaceutically acceptable for administration to a human, wherein the amino acid sequence of said mutant is SEQ ID NO:7, 8 or 9, and wherein the amino acid sequence of said fragment is SEQ ID NO:16 or 17.

63. (Previously presented) The composition of claim 62, wherein said isolated Tat protein, fragment or mutant is purified.

64. (Canceled)

65. (Previously presented) The composition of claim 62, wherein said isolated Tat protein, fragment or mutant is a wild type Tat protein.

66. (Previously presented) The composition of claim 62, 63 or 65, wherein the administration is selected from the group consisting of mucosal, nasal, oral, vaginal, rectal, intramuscular, subcutaneous, intradermal, systemic, and local administration.

67. (Canceled)

68. (Previously presented) The composition of claim 63, wherein said isolated Tat protein, fragment or mutant is purified by a method comprising performing heparin affinity chromatography.

69. (Previously presented) The composition of claim 68, wherein said performing step is followed by steps of (a) lyophilizing said isolated Tat protein, fragment or mutant, and (b) resuspending said lyophilized isolated Tat protein, fragment or mutant in a degassed buffer.

70-88. (Canceled).

89. (Previously presented) The composition of claim 62, 63 or 65 which further comprises a biologically acceptable fluid.

90. (Previously presented) A product which is produced by a process comprising lyophilizing the composition of claim 62, 63 or 65.

91. (Previously presented) A product which is produced by a process comprising lyophilizing the composition of claim 62, 63 or 65 and resuspending the lyophilized composition in a biologically acceptable fluid.

92. (Previously presented) The composition of claim 65, wherein the amino acid sequence of said wild type Tat protein consists of SEQ ID. No. 2.

93. (Previously presented) The composition of claim 89, wherein the biologically acceptable fluid is serum, plasma, or one or more fractions thereof.

94. (Previously presented) The composition of claim 91, wherein the biologically acceptable fluid is serum, plasma, or one or more fractions thereof.
95. (Previously presented) The composition of claim 62, 63, 65, or 69 which further comprises an adjuvant.
96. (Previously presented) The composition of claim 95 which further comprises a biologically acceptable fluid.
97. (Previously presented) The composition of claim 95, wherein the adjuvant is RIBI, alum, or ISCOM, or a combination thereof.
98. (Previously presented) The composition of claim 62, 63 or 65, wherein said isolated Tat protein, fragment or mutant is bound to a delivery vehicle.
99. (Previously presented) The composition of claim 98, wherein said delivery vehicle is a nanoparticle.
100. (Previously presented) The composition of claim 98, wherein said delivery vehicle is an autologous erythrocyte.
101. (Previously presented) The composition of claim 66, wherein the administration is systemic.
102. (Previously presented) The composition of claim 66, wherein the administration is intradermal.
103. (Previously presented) The composition of claim 66, wherein the administration is subcutaneous.
104. (Canceled)
105. (Previously presented) The composition of claim 66, wherein the administration is mucosal.

106. (Previously presented) The composition of claim 95, wherein the administration is selected from the group consisting of mucosal, nasal, oral, vaginal, rectal, intramuscular, subcutaneous, intradermal, systemic, and local administration.

107. (Previously presented) The composition of claim 106, wherein the administration is systemic.

108. (Previously presented) The composition of claim 106, wherein the administration is intradermal.

109. (Previously presented) The composition of claim 106, wherein the administration is subcutaneous.

110. (Previously presented) The composition of claim 109 which further comprises Alum.

111. (Previously presented) The composition of claim 106, wherein the administration is mucosal

112. (Previously presented) The composition of claim 62, 63, 65, or 69, wherein said isolated Tat protein, fragment or mutant is conjugated to a T-helper peptide or T-helper universal epitope of Tetanus Toxoid.

113. (Canceled)

114. (Previously presented) The composition of claim 62, 63, 65, or 69, which further comprises HIV rev, nef or gag, or an immunogenic fragment thereof.

115. (Canceled)

116. (Previously presented) The composition of claim 62, 63, 65, or 69, which further comprises an immuno-modulant cytokine.

117. (Previously presented) The composition of claim 116, wherein said immuno-modulant cytokine is IL-12 or IL-15.

118. (Canceled)

119. (Previously presented) The composition of claim 62, 63, 65, or 69, wherein said isolated Tat protein, fragment or mutant is fused to HIV rev, nef or gag, or an immunogenic fragment thereof.

120. (Canceled)

121. (Previously presented) The composition of claim 62, 63, 65, or 69, wherein said isolated Tat protein, fragment or mutant is fused to an immuno-modulant cytokine.

122. (Previously presented) The composition of claim 121, wherein said immuno-modulant cytokine is IL-12 or IL-15.

123. (Previously presented) The composition of claim 62, 63, 65, or 69, which further comprises an inhibitor of viral replication.

124. (Withdrawn) The composition of claim 62, 63, or 69, wherein said isolated Tat protein, fragment or mutant is a Tat mutant.

125. (Withdrawn) The composition of claim 124, wherein the amino acid sequence of said Tat mutant consists of SEQ ID NO. 7, 8, or 9.

126. (Withdrawn) The composition of claim 125, wherein the amino acid sequence of said Tat mutant consists of SEQ ID NO. 7.

127. (Previously presented) The composition of claim 62, 63, or 69, wherein said isolated Tat protein, fragment or mutant is Tat fragment.

128. (Previously presented) The composition of claim 127, wherein the amino acid sequence of said Tat fragment consists of SEQ ID NO. 16 or 17.

129-141. (Canceled)

142. (Previously presented) The composition of claim 62 which comprises a combination of said isolated Tat protein, fragment and mutant.

143. (Previously presented) The composition of claim 62 or 63 which is suitable for inducing an immune response in the human to said isolated Tat protein, fragment or mutant.

144. (Previously presented) The composition of claim 65, wherein said wild type Tat protein is purified.

145. (Previously presented) The composition of claim 92, wherein said wild type Tat protein is purified.

146. (Previously presented) The composition of claim 62, wherein said isolated Tat protein, fragment or mutant is biologically active, as shown by the ability of said isolated Tat protein, fragment or mutant to

- (iii) activate virus replication when said isolated Tat protein, fragment or mutant is added to HIV-1 infected cells at a concentration of up to 1 $\mu\text{g/ml}$, which activation is determined by (a) the rescue of Tat-defective proviruses in HLM-1 cells after the addition of said isolated Tat protein, fragment or mutant, or (b) the transactivation of HIV-1 gene expression in cells transfected with a HIV-1 promoter-reporter plasmid.

147. (Previously presented) The composition of claim 146, wherein the isolated Tat protein, fragment or mutant is a wild type Tat protein.

148. (Previously presented) The composition of claim 147, wherein said wild type Tat protein is purified.

149. (Previously presented) The composition of claim 147, wherein the amino acid sequence of said wild type Tat protein consists of SEQ ID NO. 2.

150. (Previously presented) The composition of claim 149, wherein said wild type Tat protein is purified.

151. (Previously presented) The composition of claim 146, 147, 148, or 150 which further comprises an adjuvant.

152. (Previously presented) The composition of claim 146, 147, 148, or 150, wherein the administration is intradermal.

153. (Previously presented) The composition of claim 146, 147, 148, or 150, wherein the administration is subcutaneous.

154. (Previously presented) The composition of claim 153 which further comprises Alum.

155. (Previously presented) The composition of claim 62, wherein said isolated Tat protein, fragment or mutant is biologically active, as shown by the ability of said isolated Tat protein, mutant, or fragment to

- (i) become internalized by activated endothelial cells or dendritic cells, which internalization is determined by (a) incubating activated endothelial cells or dendritic cells with up to 1 $\mu\text{g/ml}$ of said isolated Tat protein, fragment or mutant which is labeled with rhodamine, and (b) detecting the presence or absence of rhodamine in the activated endothelial cells or dendritic cells by fluorescence microscopy; and
- (ii) activate the proliferation, migration, and invasion of Kaposi's sarcoma (KS) cells or cytokine-activated endothelial cells in culture when said isolated Tat protein, fragment or mutant is present at a concentration of up to 1 $\mu\text{g/ml}$; and
- (iii) activate virus replication when said isolated Tat protein, fragment or mutant is added to HIV-1 infected cells at a concentration of up to 1 $\mu\text{g/ml}$, which activation is determined by (a) the rescue of Tat-defective proviruses in HLM-1 cells after the addition of said isolated Tat protein, fragment or mutant, or (b) the transactivation of HIV-1 gene expression in cells transfected with a HIV-1 promoter-reporter plasmid.

156. (Previously presented) The composition of claim 155, wherein the isolated Tat protein, fragment or mutant is a wild type Tat protein.

157. (Previously presented) The composition of claim 156, wherein said wild type Tat protein is purified.

158. (Previously presented) The composition of claim 156, wherein the amino acid sequence of said wild type Tat protein consists of SEQ ID NO. 2.

159. (Previously presented) The composition of claim 158, wherein said wild type Tat protein is purified.

160. (Previously presented) The composition of claim 155, 156, 157, or 159 which further comprises an adjuvant.

161. (Previously presented) The composition of claim 155, 156, 157, or 159, wherein the administration is intradermal.

162. (Previously presented) The composition of claim 155, 156, 157, or 159, wherein the administration is subcutaneous.

163. (Previously presented) The composition of claim 162 which further comprises Alum.

164. (Previously presented) The composition of claim 62 or 63, wherein said isolated Tat protein, fragment or mutant comprises the cysteine rich region of Tat and is in a non-oxidated form.

165. (Previously presented) The composition of claim 92, wherein said wild type Tat protein is in a non-oxidated form.

166. (Previously presented) The composition of claim 126, wherein said Tat mutant is in a non-oxidated form.

167. (Previously presented) The composition of any of claims 147-150, wherein said wild type Tat protein is in a non-oxidated form.

168. (Previously presented) The composition of any of claims 156-159, wherein said wild type Tat protein is in a non-oxidated form.

169. (Withdrawn) The composition of claim 146, wherein the isolated Tat protein, fragment or mutant is a Tat mutant.

170. (Withdrawn) The composition of claim 169, wherein said Tat mutant is purified.

171. (Withdrawn) The composition of claim 169, wherein the amino acid sequence of said Tat mutant consists of SEQ ID NO. 7.

172. (Withdrawn) The composition of claim 171, wherein said Tat mutant is purified.

173. (Withdrawn) The composition of any of claims 169-172, wherein said Tat mutant is in a non-oxidated form.

174. (Withdrawn) The composition of claim 155, wherein the isolated Tat protein, fragment or mutant is a Tat mutant.

175. (Withdrawn) The composition of claim 174, wherein said Tat mutant is purified.

176. (Withdrawn) The composition of claim 174, wherein the amino acid sequence of said Tat mutant consists of SEQ ID NO. 7.

177. (Withdrawn) The composition of claim 176, wherein said Tat mutant is purified.

178. (Withdrawn) The composition of any of claims 174-177, wherein said Tat mutant is in a non-oxidated form.

179. (Previously presented) A composition comprising: an isolated Tat protein, fragment or mutant in combination with a pharmaceutically acceptable carrier or excipient, wherein said isolated Tat protein, fragment or mutant comprises the cysteine rich region of Tat and is in a non-oxidated form, wherein said composition is pharmaceutically acceptable for administration to a human, wherein the amino acid sequence of said mutant is SEQ ID NO:7, 8 or 9, and wherein the amino acid sequence of said fragment is SEQ ID NO:16 or 17.

180. (Previously presented) The composition of claim 179, wherein the isolated Tat protein, fragment or mutant is a wild type Tat protein.

181. (Previously presented) The composition of claim 180, wherein said wild type Tat protein is purified.

182. (Previously presented) The composition of claim 180, wherein the amino acid sequence of said wild type Tat protein consists of SEQ ID NO. 2.

183. (Previously presented) The composition of claim 182, wherein said wild type Tat protein is purified.

184. (Previously presented) The composition of claim 179, 180, 181, or 183 which further comprises an adjuvant.

185. (Previously presented) The composition of claim 179, 180, 181, or 183, wherein the administration is intradermal.

186. (Previously presented) The composition of claim 179, 180, 181, or 183, wherein the administration is subcutaneous.

187. (Previously presented) The composition of claim 186 which further comprises Alum.

188. (Previously presented) The composition of claim 179, wherein said isolated Tat protein, fragment or mutant is conjugated to a T-helper peptide or T-helper universal epitope of Tetanus Toxoid.

189. (Previously presented) The composition of claim 179, which further comprises HIV rev, nef or gag, or an immunogenic fragment thereof.

190. (Previously presented) The composition of claim 179, which further comprises an immuno-modulant cytokine.

191. (Previously presented) The composition of claim 190, wherein said immuno-modulant cytokine is IL-12 or IL-15.

192. (Withdrawn) A composition comprising an isolated Tat mutant in combination with a pharmaceutically acceptable carrier or excipient, wherein said isolated Tat mutant is in a non-oxidated form, wherein the amino acid sequence of said Tat mutant is SEQ ID NO. 7, and wherein said composition is pharmaceutically acceptable for administration to a human.

193. (New) A method for inducing an immune response in a subject, comprising administering to said subject a composition comprising an isolated Tat protein, fragment or mutant in combination with a pharmaceutically acceptable carrier or excipient, wherein said isolated Tat protein, fragment or mutant is biologically active, as shown by the ability of said isolated Tat protein, fragment or mutant to

- (i) become internalized by activated endothelial cells or dendritic cells, which internalization is determined by (a) incubating activated endothelial cells or dendritic cells with up to 1 $\mu\text{g/ml}$ of said isolated Tat protein, fragment or mutant which is labeled with rhodamine, and (b) detecting the presence or

absence of rhodamine in the activated endothelial cells or dendritic cells by fluorescence microscopy; or

- (ii) activate the proliferation, migration, and invasion of Kaposi's sarcoma (KS) cells or cytokine-activated endothelial cells in culture when said Tat protein, fragment or mutant is present at a concentration of up to 1 µg/ml; or
- (iii) activate virus replication when said isolated Tat protein, fragment or mutant is added to HIV-1 infected cells at a concentration of up to 1 µg/ml, which activation is determined by (a) the rescue of Tat-defective proviruses in HLM-1 cells after the addition of said isolated Tat protein, fragment or mutant, or (b) the transactivation of HIV-1 gene expression in cells transfected with HIV-1 promoter-reporter plasmid,

wherein said composition is pharmaceutically acceptable for administration to a human, wherein the amino acid sequence of said mutant is SEQ ID NO:7, 8 or 9, and wherein the amino acid sequence of said fragment is SEQ ID NO:16 or 17.

194. (New) The method of claim 193, wherein said isolated Tat protein, fragment or mutant is purified.

195. (New) The method of claim 193 or 194, wherein the isolated Tat protein, fragment or mutant is a wild type Tat protein.

196. (New) A method for inducing an immune response in a subject, comprising administering to said subject a composition comprising an isolated Tat protein, fragment or mutant in combination with a pharmaceutically acceptable carrier or excipient, wherein said isolated Tat protein, fragment or mutant is in a non-oxidated form, wherein said composition is pharmaceutically acceptable for administration to a human, wherein the amino acid sequence of said mutant is SEQ ID NO:7, 8 or 9, and wherein the amino acid sequence of said fragment is SEQ ID NO:16 or 17.

197. (New) The method of claim 196, wherein said isolated Tat protein, fragment or mutant is a wild type Tat protein.

198. (New) The method of claim 196 or 197, wherein said isolated Tat protein, fragment or mutant is purified.